

CLAIM AMENDMENTS

Claims 1 through 18 (canceled)

1 19. (Currently amended) An isolated nucleic acid
2 sequence from the ATI region of modified vaccinia Ankara virus that
3 ~~includes at least one restriction enzyme recognition site as an~~
4 ~~insertion site for a heterologous sequence and that hybridizes~~
5 under stringent conditions to the nucleic acid sequence of SEQ ID
6 NO:1 or its complementary strand which includes multiple cloning
7 sites inserted into an open reading frame or the ECORI site of the
8 isolated nucleic acid sequence, said nucleic acid sequence capable
9 of integration of ~~[[the]]~~ a heterologous sequence through
10 homologous recombination into an open reading frame or an ECORI
11 site of the ATI region of an orthopoxvirus without interfering with
12 its viral propagation or replication efficiency.

1 20. (Currently amended) The nucleic acid sequence
2 defined in claim 19 that includes as the insertion site for the
3 multiple cloning sites an ECORI site corresponding to position 1063
4 of SEQ ID NO:1 that hybridizes under stringent conditions to the
5 nucleic acid sequence of SEQ ID NO:1 or its complementary strand.

1 21. (Currently amended) An isolated fragment of a
2 nucleic acid sequence from the ATI region of modified vaccinia
3 Ankara virus consisting essentially of at least 200 base pairs of

4 the nucleic acid sequence that is SEQ ID NO:1, ~~that is at least 70%~~
5 ~~homologous to SEQ ID NO:1 and that includes at least one~~
6 ~~restriction enzyme recognition site as an insertion site for a~~
7 ~~heterologous sequence, which includes multiple cloning sites~~
8 inserted into an open reading frame or the ECORI site of the
9 isolated fragment of a nucleic acid sequence, said isolated
10 fragment of a nucleic acid sequence capable of integration of
11 [[the]] a heterologous sequence through homologous recombination
12 into an open reading frame or an ECORI site of the ATI region of an
13 orthopoxvirus without interfering with its viral propagation or
14 replication efficiency.

1 22. (Currently amended) The isolated fragment of a
2 nucleic acid sequence defined in claim 21 that includes as the
3 insertion site for the multiple cloning sites an ECORI site
4 corresponding to position 1063 of SEQ ID NO:1.

1 23. (Currently amended) A vector for integration of a
2 heterologous sequence into an open reading frame of or into the
3 ECORI site of the ATI region of an orthopoxviral genome having an
4 ATI region, said vector including an isolated nucleic acid sequence
5 from the ATI region of modified vaccinia Ankara virus ~~that includes~~
6 ~~at least one restriction enzyme recognition site as an insertion~~
7 ~~site for a heterologous nucleic acid sequence, that hybridizes~~
8 under stringent conditions to the nucleic acid sequence of SEQ ID
9 NO:1 or its complementary strand, which includes multiple cloning

10 sites inserted into an open reading frame or the ECORI site of the
11 isolated nucleic acid sequence, and that is said nucleic acid
12 sequence and that is capable of integration of the heterologous
13 sequence through homologous recombination into an open reading
14 frame or the ECORI site of the ATI region of an orthopoxvirus
15 without interfering with its viral propagation or replication
16 efficiency.

1 24. (Previously presented) The vector defined in claim
2 23 wherein additionally at least one transcriptional control
3 element is included in the insertion site.

1 25. (Previously presented) The vector defined in claim
2 23 wherein the insertion site is the restriction site ECOR1.

1 26. (Previously presented) The vector defined in claim
2 24 wherein the at least one transcriptional control element is
3 obtained from a poxvirus genome or is a consensus sequence from a
4 poxvirus genome.

1 27. (Currently amended) The vector defined in claim 23
2 further comprising at least one heterologous sequence inserted
3 within the insertion site into an open reading frame or the ECORI
4 site of the isolated nucleic acid sequence, said heterologous
5 nucleic acid sequence functionally associated with a
6 transcriptional control element thereof.

1 28. (Previously presented) The vector defined in claim
2 27 wherein the heterologous nucleic sequence is selected from the
3 group consisting of marker genes, therapeutic genes, host range
4 genes and genes encoding immunogenic epitopes.

1 29. (Previously presented) The vector defined in claim
2 27 comprising a recombinogenic sequence, which flanks one or more
3 heterologous sequences encoding marker genes, host range genes,
4 and/or a transcriptional element thereof.

1 30. (Currently amended) A vector for integration of a
2 heterologous sequence into an open reading frame of or into the
3 ECORI site of the ATI region of an orthopoxviral genome having an
4 ATI region, said vector including an isolated fragment of a nucleic
5 acid sequence from the ATI region of modified vaccinia Ankara virus
6 consisting essentially of at least 200 base pairs of the nucleic
7 acid sequence that is SEQ ID NO:1, ~~that is at least 70% homologous~~
8 ~~to SEQ ID NO:1 and that includes at least one restriction enzyme~~
9 ~~recognition site as an insertion site for the heterologous~~
10 ~~sequence, which includes multiple cloning sites inserted into an~~
11 open reading frame or the ECORI site of the isolated fragment of a
12 nucleic acid sequence, said isolated fragment of the nucleic acid
13 sequence capable of integration of the heterologous sequence
14 through homologous recombination into an open reading frame or an

15 ECORI site of the ATI region of an orthopoxvirus without
16 interfering with its viral propagation or replication efficiency.

1 31. (Previously presented) The vector defined in claim
2 30 wherein additionally at least one transcriptional control
3 element is included in the insertion site.

1 32. (Previously presented) The vector defined in claim
2 30 wherein the insertion site is the restriction site ECOR1.

1 33. (Previously presented) The vector defined in claim
2 31 wherein the at least one transcriptional control element is
3 obtained from a poxvirus genome or is a consensus sequence from a
4 poxvirus genome.

1 34. (Currently amended) The vector defined in claim 30
2 further comprising at least one heterologous sequence inserted
3 within the insertion site into an open reading frame or the ECORI
4 site of the isolated nucleic acid sequence,, said heterologous
5 nucleic acid sequence functionally associated with a
6 transcriptional control element thereof.

1 35. (Previously presented) The vector defined in claim
2 34 wherein the heterologous sequence is selected from the group
3 consisting of marker genes, therapeutic genes, host range genes and
4 genes encoding immunogenic epitopes.

1 36. (Previously presented) The vector defined in claim
2 34 comprising a recombinogenic nucleic acid sequence, which flanks
3 one or more heterologous sequences encoding marker genes, host
4 range genes, and/or a transcriptional element thereof.

1 37. (Currently amended) A recombinant orthopoxvirus
2 having an ATI region, comprising in an open reading frame of or in
3 the ECORI site of its ATI region an integrated heterologous nucleic
4 acid sequence wherein said integrated heterologous nucleic acid
5 sequence does not interfere with viral propagation and/or
6 replication efficiency.

1 38. (Previously presented) The recombinant
2 orthopoxvirus defined in claim 37 wherein the orthopoxvirus is
3 selected from the group consisting of a modified vaccinia Ankara
4 virus, vaccinia virus Western Reserve, and vaccinia virus
5 Copenhagen.

1 39. (Previously presented) The recombinant
2 orthopoxvirus defined in claim 37 wherein the orthopoxvirus is the
3 modified vaccinia Ankara virus.

1 40. (Previously presented) The recombinant
2 orthopoxvirus defined in claim 37 wherein the heterologous sequence

3 integrated into the orthopoxvirus in its ATI region is from the ATI
4 region of modified vaccinia Ankara virus.

1 41. (Previously presented) The recombinant
2 orthopoxvirus defined in claim 40 wherein the orthopoxvirus is
3 selected from the group consisting of a modified vaccinia Ankara
4 virus, vaccinia virus Western Reserve, and vaccinia virus
5 Copenhagen.

1 42. (Previously presented) The recombinant
2 orthopoxvirus defined in claim 40 wherein the orthopoxvirus is the
3 modified vaccinia Ankara virus.

1 43. (Currently amended) A recombinant orthopoxvirus
2 comprising an ATI region including within an open reading frame of
3 or within the ECORI site of the ATI region an integrated
4 heterologous sequence wherein said recombinant orthopoxvirus is
5 obtained by a method comprising the steps of:

6 (a) transducing a host cell with a vector which comprises
7 an isolated nucleic acid sequence from the ATI region of modified
8 vaccinia Ankara virus ~~that includes at least one restriction enzyme~~
9 ~~recognition site as an insertion site for the heterologous~~
10 ~~sequence,~~ that hybridizes under stringent conditions to the nucleic
11 acid sequence of SEQ ID NO:1 or its complementary strand, ~~and that~~
12 ~~is said nucleic acid sequence~~ capable of integration of the
13 heterologous sequence through homologous recombination into an open

14 reading frame or the ECORI site of the ATI region of an
15 orthopoxvirus without interfering with its viral propagation or
16 replication efficiency, and at least one heterologous sequence
17 inserted within the insertion site into an open reading frame or
18 the ECORI site of the isolated nucleic acid sequence;

19 (b) infecting said host cell with an orthopoxvirus having
20 an ATI region;

21 (c) integrating the heterologous sequence into an open
22 reading frame or the ECORI site of the ATI region of the
23 orthopoxvirus by homologous recombination between the nucleic acid
24 sequence and a corresponding genomic sequence of the orthopoxvirus
25 to obtain a recombinant orthopoxvirus; and

26 (d) isolating said recombinant orthopoxvirus.

1 44. (Currently amended) A recombinant orthopoxvirus
2 comprising an ATI region including within an open reading frame of
3 or within the ECORI site of the ATI region an integrated
4 heterologous sequence wherein said recombinant orthopoxvirus is
5 obtained by a method comprising the steps of:

6 (a) transducing a host cell with a vector which comprises
7 an isolated fragment of a nucleic acid sequence from the ATI region
8 of modified vaccinia Ankara virus consisting essentially of at
9 least 200 base pairs of the nucleic acid sequence that is SEQ ID
10 NO:1, ~~that is at least 70% homologous to SEQ ID NO:1 and that~~
11 ~~includes at least one restriction enzyme recognition site as an~~
12 ~~insertion site for the heterologous sequence,~~ said isolated

13 fragment of the nucleic acid sequence capable of integration of the
14 heterologous sequence through homologous recombination into an open
15 reading frame or an ECORI site of the ATI region of an
16 orthopoxvirus without interfering with its viral propagation or
17 replication efficiency and at least one heterologous sequence
18 inserted within the insertion site into an open reading frame or
19 the ECORI site of the isolated nucleic acid sequence;

20 (b) infecting said host cell with an orthopoxvirus having
21 an ATI region;

22 (c) integrating the heterologous sequence into an open
23 reading frame or the ECORI site of the ATI region of the
24 orthopoxvirus by homologous recombination between the isolated
25 fragment of the nucleic acid sequence and a corresponding genomic
26 sequence of the orthopoxvirus to obtain a recombinant
27 orthopoxvirus; and

28 (d) isolating said recombinant orthopoxvirus.

1 45. (Currently amended) A recombinant orthopoxvirus
2 comprising an ATI region including within an open reading frame of
3 or within the ECORI site of the ATI region [[a]] an integrated
4 heterologous sequence wherein said recombinant orthopoxvirus is
5 obtained by a method comprising the steps of:

6 (a) transducing a host cell with a vector which comprises
7 an isolated nucleic acid sequence according to SEQ ID NO:1 or its
8 complementary strand from the ATI region of modified vaccinia
9 Ankara virus ~~that includes at least one restriction enzyme~~

10 ~~recognition site as an insertion site for the heterologous~~
11 ~~sequence, and that is capable of integration of the heterologous~~
12 ~~sequence into an open reading frame or an ECORI site of the ATI~~
13 ~~region of an orthopoxvirus without interfering with its viral~~
14 ~~propagation or replication efficiency, and at least one~~
15 ~~heterologous sequence inserted within the insertion site into an~~
16 ~~open reading frame or the ECORI site of the isolated nucleic acid~~
17 ~~sequence;~~

18 (b) infecting said host cell with an orthopoxvirus having
19 an ATI region;

20 (c) integrating the heterologous sequence into an open
21 reading frame or the ECORI site of the ATI region of the
22 orthopoxvirus by homologous recombination between the nucleic acid
23 sequence and a corresponding genomic sequence of the orthopoxvirus
24 to obtain a recombinant orthopoxvirus; and

25 (d) isolating said recombinant orthopoxvirus.

1 46. (Currently amended) A method of integrating a
2 heterologous sequence into an open reading frame or the ECORI site
3 of the ATI region of an orthopoxvirus to obtain a recombinant
4 orthopoxvirus which comprises the steps of:

5 (a) transducing a host cell with a vector comprising an
6 isolated nucleic acid sequence from the ATI region of modified
7 vaccinia Ankara virus ~~that includes at least one restriction enzyme~~
8 ~~recognition site as an insertion site for the heterologous~~
9 ~~sequence,~~ that hybridizes under stringent conditions to the nucleic

10 acid sequence of SEQ ID NO:1 or its complementary strand, and that
11 is said nucleic acid sequence capable of integration of the
12 heterologous sequence through homologous recombination into an open
13 reading frame or the ECORI site of the ATI region of an
14 orthopoxvirus without interfering with its viral propagation and
15 replication efficiency, and at least one heterologous sequence
16 inserted within the insertion site into an open reading frame or
17 the ECORI site of the isolated nucleic acid sequence;

18 (b) infecting said host cell with an orthopoxvirus having
19 an ATI region;

20 (c) integrating the heterologous sequence into an open
21 reading frame or the ECORI site of the ATI region of the
22 orthopoxvirus by homologous recombination between the nucleic acid
23 sequence and a corresponding genomic sequence of the orthopoxvirus
24 to obtain a recombinant orthopoxvirus; and

25 (d) isolating said recombinant orthopoxvirus.

1 47. (Currently amended) The method of integrating a
2 heterologous sequence into the open reading frame or the ECORI site
3 of the ATI region of the orthopoxvirus defined in claim 46 wherein
4 according to step (b) the orthopoxvirus is modified vaccinia Ankara
5 virus.

1 48. (Currently amended) A method of integrating a
2 heterologous sequence into an open reading frame or the ECORI site

3 of the ATI region of an orthopoxvirus to obtain a recombinant
4 orthopoxvirus which comprises the steps of:

5 (a) transducing a host cell with a vector comprising an
6 isolated fragment of a nucleic acid sequence from the ATI region of
7 modified vaccinia Ankara virus consisting essentially of at least
8 200 base pairs of the nucleic acid sequence that is SEQ ID NO:1,
9 ~~that is at least 70% homologous to SEQ ID NO:1 and that includes at~~
10 ~~least one restriction enzyme recognition site as an insertion site~~
11 ~~for the heterologous sequence,~~ said isolated fragment of the
12 nucleic acid sequence capable of integration of the heterologous
13 sequence into the ATI region of an orthopoxvirus through homologous
14 recombination into an open reading frame or an ECORI site of the
15 ATI region without interfering with its viral propagation or
16 replication efficiency and at least one heterologous sequence
17 inserted within the insertion site;

18 (b) infecting said host cell with an orthopoxvirus having
19 an ATI region;

20 (c) integrating the heterologous sequence into an open
21 reading frame or the ECORI site of the ATI region of the
22 orthopoxvirus by homologous recombination between the isolated
23 fragment of the nucleic acid sequence and a corresponding genomic
24 sequence of the orthopoxvirus to obtain a recombinant
25 orthopoxvirus; and

26 (d) isolating said recombinant orthopoxvirus.

1 49. (Currently amended) The method of integrating a
2 heterologous sequence into an open reading frame or the ECORI site
3 of the ATI region of the orthopoxvirus defined in claim 48 wherein
4 according to step (b) the orthopoxvirus is modified vaccinia Ankara
5 virus.

1 50. (Previously presented) A target cell comprising the
2 recombinant orthopoxvirus defined in claim 37.

1 51. (Previously presented) A pharmaceutical composition
2 for effecting an immune response against an infectious disease or a
3 proliferative disorder which consists essentially of a
4 therapeutically effective amount of the recombinant orthopoxvirus
5 as defined in claim 37 and in a form capable of producing an immune
6 response against an infectious disease or a proliferative disorder
7 in combination with a pharmaceutically acceptable inert carrier or
8 diluent.

1 52. (Previously presented) A method of effecting an
2 immune response against an infectious disease or a proliferative
3 disorder in an animal subject which comprises the step of
4 administering to said subject a therapeutically effective amount of
5 the pharmaceutical composition defined in claim 51.

1 53. (New) A vector for integration of a heterologous
2 sequence into an open reading frame of or into the ECORI site of

3 the ATI region of an orthopoxviral genome having an ATI region,
4 said vector including an isolated nucleic acid sequence from the
5 ATI region of modified vaccinia Ankara virus, that hybridizes under
6 stringent conditions to the nucleic acid sequence of SEQ ID NO:1 or
7 its complementary strand, which includes at least one heterologous
8 sequence inserted into an open reading frame or the ECORI site of
9 the isolated nucleic acid sequence, said at least one heterologous
10 sequence functionally associated with a transcriptional control
11 element thereof, said isolated nucleic acid sequence capable of
12 integration of the heterologous sequence through homologous
13 recombination into an open reading frame or the ECORI site of the
14 ATI region of an orthopoxvirus without interfering with its viral
15 propagation or replication efficiency.

1 54. (New) A vector for integration of a heterologous
2 sequence into an open reading frame of or into the ECORI site of
3 the ATI region of an orthopoxviral genome having an ATI region,
4 said vector including an isolated fragment of a nucleic acid
5 sequence from the ATI region of modified vaccinia Ankara virus
6 consisting essentially of at least 200 base pairs of the nucleic
7 acid sequence that is SEQ ID NO:1, which includes at least one
8 heterologous sequence inserted into an open reading frame or the
9 ECORI site of the isolated fragment of a nucleic acid sequence, and
10 which is functionally associated with a transcriptional control
11 element thereof, said isolated fragment of the nucleic acid
12 sequence capable of integration of the heterologous sequence

13 through homologous recombination into an open reading frame or an
14 ECORI site of the ATI region of an orthopoxvirus without
15 interfering with its viral propagation or replication efficiency.